

***Remarks***

Upon entry of the foregoing amendment, claims 1-37 are pending in the application, with claims 1, 14, 25 and 27 being the independent claims. Claims 38 and 39 are sought to be cancelled without prejudice to or disclaimer of the subject matter therein. Claims 3-13, 16-24, 29-31 and 33-37 were amended only to eliminate multiple dependencies and correct spelling errors. Hence, no new matter has been added by the amendment and entry and consideration of the same is respectfully requested.

***Conclusion***

It is respectfully believed that the present application is in condition for examination. Early notice to this effect is earnestly solicited. If the Examiner believes, for any reason, that personal communication will expedite prosecution of this application, the Examiner is invited to telephone the undersigned at the number provided.

Respectfully submitted,

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**Version with markings to show changes made**

***In the Claims:***

The pending claims 3-13, 16-24, 29-31 and 33-37 were substituted by the following claims 3-13, 16-24, 29-31 and 33-37.

3. (Once amended) Method according to claim 1 [or 2], characterized [characterised] in that the prokaryotic cell is *E. coli*.
4. (Once amended) Method according to [one of claims] claim 1 [to 3], characterized [characterised] in that the [the] following steps are carried out:
  - a) the DNA encoding the tPA, tPA variant, K2S molecule or K2S variant is amplified by PCR;
  - b) the PCR product is purified;
  - c) said PCR product is inserted into a vector comprising the DNA coding for OmpA signal peptide and the DNA coding for gpIII in such a way that said PCR product is operably linked upstream to the DNA coding for the OmpA signal sequence and linked downstream to the DNA coding for gpIII of said vector;
  - d) that a stop codon is inserted between said tPA, tPA variant, K2S molecule or K2S variant and gpIII;
  - e) said vector is expressed by the prokaryotic cell;
  - f) the tPA, tPA variant, K2S molecule or K2S variant is purified.

5. (Once amended) Method according to [one of claims] claim 1 [to 4],  
characterized [characterised] in that the vector is a phagemid vector comprising the DNA  
coding for OmpA signal peptide and the DNA coding for gpIII.

6. (Once amended) Method according to [one of claims] claim 1 [to 5],  
characterized [characterised] in that the vector is the pComb3HSS phagemid.

7. (Once amended) Method according to [one of claims] claim 1 [to 6],  
characterized [characterised] in that the DNA Sequence of OmpA linked upstream to  
K2S comprises the following sequence or a functional variant thereof or a variant due to  
the degenerate nucleotide code:

ATGAAAAAGACAGCTATCGCGATTGCAGTGGCACTGGCTGGTTTCGCTACCG  
TGGCCCAGGCGGCCTCTGAGGGAAACAGTGACTGCTACTTTGGGAATGGGTC  
AGCCTACCGTGGCACGCACAGCCTCACCGAGTCGGGTGCCTCCTGCCTCCCG  
TGGAATTCCATGATCCTGATAGGCAAGGTTTACACAGCACAGAAACCCAGTG  
CCCAGGCACTGGGCCTGGGCAAACATAATTACTGCCGGAATCCTGATGGGGA  
TGCCAAGCCCTGGTGCCACGTGCTGAAGAACCGCAGGCTGACGTGGGAGTA  
CTGTGATGTGCCCTCCTGCTCCACCTGCGGCCTGAGACAGTACAGCCAGCCT  
CAGTTTCGCATCAAAGGAGGGCTCTTCGCCGACATCGCCTCCCACCCCTGGC  
AGGCTGCCATCTTTGCCAAGCACAGGAGGTCGCCCCGAGAGCGGTTCTGTG  
CGGGGGCATACTCATCAGCTCCTGCTGGATTCTCTCTGCCGCCCCACTGCTTCC

AGGAGAGGTTTCCGCCCCACCACCTGACGGTGATCTTGGGCAGAACATACCG  
GGTGGTCCCTGGCGAGGAGGAGCAGAAATTTGAAGTCGAAAAATACATTGT  
CCATAAGGAATTCGATGATGACACTTACGACAATGACATTGCGCTGCTGCAG  
CTGAAATCGGATTCGTCCCGCTGTGCCCAGGAGAGCAGCGTGGTCCGCACTG  
TGTGCCTTCCCCCGGCGGACCTGCAGCTGCCGGACTGGACGGAGTGTGAGCT  
CTCCGGCTACGGCAAGCATGAGGCCTTGTCTCCTTTCTATTCGGAGCGGCTG  
AAGGAGGCTCATGTCAGACTGTACCCATCCAGCCGCTGCACATCACAACATT  
TACTTAACAGAACAGTCACCGACAACATGCTGTGTGCTGGAGACACTCGGAG  
CGGCGGGCCCCAGGCAAACCTTGCACGACGCCTGCCAGGGCGATTCGGGAGG  
CCCCCTGGTGTGTCTGAACGATGGCCGCATGACTTTGGTGGGCATCATCAGC  
TGGGGCCTGGGCTGTGGACAGAAGGATGTCCCGGGTGTGTACACAAAGGTTA  
CCAACTACCTAGACTGGATTCGTGACAACATGCGACCG (SEQ ID NO:2)

8. (Once amended) Method according to [one of claims] claim 1 [to 7],  
characterized [characterised] in that the DNA Sequence of OmpA comprises the  
following sequence:

ATGAAAAAGACAGCTATCGCGATTGCAGTGGCACTGGCTGGTTTCGCTACCG  
TGGCCCAGGCGGCC (SEQ ID NO:3)

9. (Once amended) Method according to [one of claims] claim 1 [to 8],  
characterized [characterised] in that the DNA Sequence of OmpA consists of the  
following sequence:

ATGAAAAAGACAGCTATCGCGATTGCAGTGGCACTGGCTGGTTTCGCTACCG  
TGGCCCAGGCGGCC (SEQ ID NO:3)

10. (Once amended) Method according to [one of claims] claim 1 [to 9],  
characterized [characterised] in that the DNA of the tPA, tPA variant, K2S molecule or  
K2S variant is preceded [preceeded] by a lac promoter [promotor] and/or a ribosomal  
binding site.
11. (Once amended) Method according to [one of claims] claim 1 [to 10],  
characterized [characterised] in that the DNA coding for the tPA, tPA variant, K2S  
molecule or K2S variant is selected from the group of DNA molecules coding for at least  
90% of the amino acids 87 - 527, 174 - 527, 180 - 527 or 220 - 527 of the human tissue  
plasminogen activator protein.
12. (Once amended) Method according to [one of claims] claim 5 [to 11],  
characterized [characterised] in that the DNA Sequence of K2S comprises the following  
sequence or a functional variant thereof or a variant due to the degenerate nucleotide  
code:
- TCTGAGGGAAACAGTGACTGCTACTTTGGGAATGGGTCAGCCTACCGTGG  
CACGCACAGCCTCACCGAGTCGGGTGCCTCCTGCCTCCCGTGGAATTCCA  
TGATCCTGATAGGCAAGGTTTACACAGCACAGAACCCAGTGCCCAGGC  
ACTGGGCCTGGGCAAACATAATTACTGCCGGAATCCTGATGGGGATGCC  
AAGCCCTGGTGCCACGTGCTGAAGAACCGCAGGCTGACGTGGGAGTACT

GTGATGTGCCCTCCTGCTCCACCTGCGGCCTGAGACAGTACAGCCAGCCT  
CAGTTTCGCATCAAAGGAGGGCTCTTCGCCGACATCGCCTCCCACCCCTG  
GCAGGCTGCCATCTTTGCCAAGCACAGGAGGTCGCCCCGAGAGCGGTTC  
CTGTGCGGGGGCATACTCATCAGCTCCTGCTGGATTCTCTCTGCCGCCCA  
CTGCTTCCAGGAGAGGTTTCCGCCCCACCACCTGACGGTGATCTTGGGCA  
GAACATAACGGGTGGTCCCTGGCGAGGAGGAGCAGAAATTTGAAGTCGA  
AAAATACATTGTCCATAAGGAATTCGATGATGACACTTACGACAATGACA  
TTGCGCTGCTGCAGCTGAAATCGGATTCGTCCCGCTGTGCCCAGGAGAGC  
AGCGTGGTCCGCACTGTGTGCCTTCCCCCGGCGGACCTGCAGCTGCCGGA  
CTGGACGGAGTGTGAGCTCTCCGGCTACGGCAAGCATGAGGCCTTGTCTC  
CTTTCTATTCGGAGCGGCTGAAGGAGGCTCATGTCAGACTGTACCCATCC  
AGCCGCTGCACATCACAACATTTACTTAACAGAACAGTCACCGACAACA  
TGCTGTGTGCTGGAGACACTCGGAGCGGCGGGCCCCAGGCCAACTTGCA  
CGACGCCTGCCAGGGCGATTTCGGGAGGCCCCCTGGTGTGTCTGAACGAT  
GGCCGCATGACTTTGGTGGGCATCATCAGCTGGGGCCTGGGCTGTGGAC  
AGAAGGATGTCCCGGGTGTGTACACAAAGGTTACCAACTACCTAGACTG  
GATTCGTGACAACATGCGACCGTGA (SEQ ID NO:4).

13. (Once amended) Method according to [one of claims] claim 5 [to 12],  
characterized [characterised] in that the DNA Sequence of K2S consists of the following  
sequence:

TCTGAGGGAAACAGTGACTGCTACTTTGGGAATGGGTCAGCCTACCGTGG  
CACGCACAGCCTCACCGAGTCGGGTGCCTCCTGCCTCCCGTGGAATTCCA

TGATCCTGATAGGCAAGGTTTACACAGCACAGAACCCCAGTGCCCAGGC  
ACTGGGCCTGGGCAAACATAATTACTGCCGGAATCCTGATGGGGATGCC  
AAGCCCTGGTGCCACGTGCTGAAGAACCGCAGGCTGACGTGGGAGTACT  
GTGATGTGCCCTCCTGCTCCACCTGCGGCCTGAGACAGTACAGCCAGCCT  
CAGTTTCGCATCAAAGGAGGGCTCTTCGCCGACATCGCCTCCCACCCCTG  
GCAGGCTGCCATCTTTGCCAAGCACAGGAGGTCGCCCCGGAGAGCGGTTC  
CTGTGCGGGGGCATACTCATCAGCTCCTGCTGGATTCTCTCTGCCGCCCA  
CTGCTTCCAGGAGAGGTTTCCGCCCCACCACCTGACGGTGATCTTGGGCA  
GAACATAACGGGTGGTCCCTGGCGAGGAGGAGCAGAAATTTGAAGTCGA  
AAAATACATTGTCCATAAGGAATTCGATGATGACACTTACGACAATGACA  
TTGCGCTGCTGCAGCTGAAATCGGATTCGTCCCGCTGTGCCCAGGAGAGC  
AGCGTGGTCCGCACTGTGTGCCTTCCCCCGGCGGACCTGCAGCTGCCGGA  
CTGGACGGAGTGTGAGCTCTCCGGCTACGGCAAGCATGAGGCCTTGTCTC  
CTTTCTATTCGGAGCGGCTGAAGGAGGCTCATGTCAGACTGTACCCATCC  
AGCCGCTGCACATCACAACATTTACTTAACAGAACAGTCACCGACAACA  
TGCTGTGTGCTGGAGACACTCGGAGCGGCGGGCCCCAGGCAAACCTTGCA  
CGACGCCTGCCAGGGCGATTCGGGAGGCCCCCTGGTGTGTCTGAACGAT  
GGCCGCATGACTTTGGTGGGCATCATCAGCTGGGGCCTGGGCTGTGGAC  
AGAAGGATGTCCCGGGTGTGTACACAAAGGTTACCAACTACCTAGACTG  
GATTCGTGACAACATGCGACCGTGA (SEQ ID NO:4).

16. (Once amended) DNA molecule according to claim 14 [or 15], characterized in that said DNA sequence consists of the following sequence:

ATGAAAAAGACAGCTATCGCGATTGCAGTGGCACTGGCTGGTTTCGCTACCG  
TGGCCCAGGCGGCCTCTGAGGGAAACAGTGACTGCTACTTTGGGAATGGGTC  
AGCCTACCGTGGCACGCACAGCCTCACCGAGTCGGGTGCCTCCTGCCTCCCG  
TGGAATTCCATGATCCTGATAGGCAAGGTTTACACAGCACAGAACCCCAAGTG  
CCCAGGCACTGGGCCTGGGCAAACATAATTACTGCCGGAATCCTGATGGGGA  
TGCCAAGCCCTGGTGCCACGTGCTGAAGAACCGCAGGCTGACGTGGGAGTA  
CTGTGATGTGCCCTCCTGCTCCACCTGCGGCCTGAGACAGTACAGCCAGCCT  
CAGTTTCGCATCAAAGGAGGGCTCTTCGCCGACATCGCCTCCCACCCCTGGC  
AGGCTGCCATCTTTGCCAAGCACAGGAGGTCGCCCCGAGAGCGGTTCTGT  
GCGGGGGCATACTCATCAGCTCCTGCTGGATTCTCTCTGCCGCCCCTGCTT  
CCAGGAGAGGTTTCCGCCCCACCACCTGACGGTGATCTTGGGCAGAACATA  
CCGGGTGGTCCCTGGCGAGGAGGAGCAGAAATTTGAAGTCGAAAAATACA  
TTGTCCATAAGGAATTCGATGATGACACTTACGACAATGACATTGCGCTGC  
TGCAGCTGAAATCGGATTCGTCCCGCTGTGCCCAGGAGAGCAGCGTGGTCC  
GCACTGTGTGCCTTCCCCCGGCGGACCTGCAGCTGCCGGACTGGACGGAGT  
GTGAGCTCTCCGGCTACGGCAAGCATGAGGCCTTGTCTCCTTTCTATTCGGA  
GCGGCTGAAGGAGGCTCATGTCAGACTGTACCCATCCAGCCGCTGCACATC  
ACAACATTTACTTAACAGAACAGTCACCGACAACATGCTGTGTGCTGGAGAC  
ACTCGGAGCGGCGGGCCCCAGGCAAACCTTGACGACGCCTGCCAGGGCGAT  
TCGGGAGGCCCCCTGGTGTGTCTGAACGATGGCCGCATGACTTTGGTGGGCA  
TCATCAGCTGGGGCCTGGGCTGTGGACAGAAGGATGTCCCGGGTGTGTACAC  
AAAGGTTACCAACTACCTAGACTGGATTTCGTGACAACATGCGACCG (SEQ ID  
NO:5).



17. (Once amended) DNA molecule according to [one of claims] claim 14 [to 16], characterized in that said DNA sequence b) is coding for at least 90% of the amino acids 87 - 527 of the human tissue plasminogen activator protein.

18. (Once amended) DNA molecule according to [one of claims] claim 14 [to 17], characterized in that said DNA sequence b) is coding for at least 90% of the amino acids 174 - 527 of the human tissue plasminogen activator protein.

19. (Once amended) DNA molecule according to [any one of claims] claim 14 [to 18], characterized in that said DNA sequence b) is coding for at least 90% of the amino acids 180 - 527 of the human tissue plasminogen activator protein.

20. (Once amended) DNA molecule according to [any one of claims] claim 14 [to 19], characterized in that said DNA sequence b) is coding for at least 90% of the amino acids 220 - 527 of the human tissue plasminogen activator protein.

21. (Once amended) DNA molecule according to [any one of claims] claim 14 [to 20], characterized in that said DNA sequence a) is hybridizing under stringent conditions to the following sequence:

ATGAAAAAGACAGCTATCGCGATTGCAGTGGCACTGGCTGGTTTCGCTACCG  
TGGCCCAGGCGGCC (SEQ ID NO:6).

22. (Once amended) DNA molecule according to [any one of claims] claim 14 [to 21], characterized in that said DNA sequence a) consists of the following sequence:

ATGAAAAAGACAGCTATCGCGATTGCAGTGGCACTGGCTGGTTTCGCTACCG  
TGGCCCAGGCGGCC (SEQ ID NO:6).

23. (Once amended) DNA molecule according to [any one of claims] claim 14 [to 22], characterized in that said DNA sequence b) is hybridizing under stringent conditions to the following sequence:

TCTGAGGGAAACAGTGACTGCTACTTTGGGAATGGGTCAGCCTACCGTGGC  
ACGCACAGCCTCACCGAGTCGGGTGCCTCCTGCCTCCCGTGGAATTCCATG  
ATCCTGATAGGCAAGGTTTACACAGCACAGAACCCCAGTGCCCAGGCACT  
GGGCCTGGGCAAACATAATTACTGCCGGAATCCTGATGGGGATGCCAAGC  
CCTGGTGCCACGTGCTGAAGAACCGCAGGCTGACGTGGGAGTACTGTGAT  
GTGCCCTCCTGCTCCACCTGCGGCCTGAGACAGTACAGCCAGCCTCAGTTT  
CGCATCAAAGGAGGGCTCTTCGCCGACATCGCCTCCCACCCCTGGCAGGCT  
GCCATCTTTGCCAAGCACAGGAGGTCGCCCCGAGAGCGGTTCTGTGCGGG  
GGCATACTCATCAGCTCCTGCTGGATTCTCTCTGCCGCCCCACTGCTTCCAGG  
AGAGGTTTCCGCCCCACCACCTGACGGTGATCTTGGGCAGAACATAACGGG  
TGGTCCCTGGCGAGGAGGAGCAGAAATTTGAAGTCGAAAAATACATTGTCC  
ATAAGGAATTCGATGATGACACTTACGACAATGACATTGCGCTGCTGCAGCT  
GAAATCGGATTCGTCCCGCTGTGCCCAGGAGAGCAGCGTGGTCCGCACTGTG  
TGCCTTCCCCCGGCGGACCTGCAGCTGCCGGACTGGACGGAGTGTGAGCTCT  
CCGGCTACGGCAAGCATGAGGCCTTGTCTCCTTTCTATTCCGAGCGGCTGAA

GGAGGCTCATGTCAGACTGTACCCATCCAGCCGCTGCACATCACAACATTTA  
CTTAACAGAACAGTCACCGACAACATGCTGTGTGCTGGAGACACTCGGAGCG  
GCGGGCCCCAGGCAAACCTTGACGACGCCTGCCAGGGCGATTCTGGGAGGCC  
CCCTGGTGTGTCTGAACGATGGCCGCATGACTTTGGTGGGCATCATCAGCTG  
GGGCCTGGGCTGTGGACAGAAGGATGTCCCGGGTGTGTACACAAAGGTTAC  
CAACTACCTAGACTGGATTCGTGACAACATGCGACCGTGA (SEQ ID NO:7).

24. (Once amended) DNA molecule according to [any one of claims] claim 14 [to  
23], characterized in that said DNA sequence b) consists of the following sequence:

TCTGAGGGAAACAGTGACTGCTACTTTGGGAATGGGTCAGCCTACCGTGGC  
ACGCACAGCCTCACCGAGTCGGGTGCCTCCTGCCTCCCGTGGAATTCCATG  
ATCCTGATAGGCAAGGTTTACACAGCACAGAACCCCAGTGCCCAGGCACT  
GGGCCTGGGCAAACATAATTACTGCCGGAATCCTGATGGGGATGCCAAGC  
CCTGGTGCCACGTGCTGAAGAACCGCAGGCTGACGTGGGAGTACTGTGAT  
GTGCCCTCCTGCTCCACCTGCGGCCTGAGACAGTACAGCCAGCCTCAGTTT  
CGCATCAAAGGAGGGCTCTTCGCCGACATCGCCTCCCACCCCTGGCAGGCT  
GCCATCTTTGCCAAGCACAGGAGGTCGCCCCGAGAGCGGTTCTGTGCGGG  
GGCATACTCATCAGCTCCTGCTGGATTCTCTCTGCCGCCCACTGCTTCCAGG  
AGAGGTTTCCGCCCCACCACCTGACGGTGATCTTGGGCAGAACATACCGGG  
TGGTCCCTGGCGAGGAGGAGCAGAAATTTGAAGTCGAAAAATACATTGTCC  
ATAAGGAATTCGATGATGACACTTACGACAATGACATTGCGCTGCTGCAGC  
TGAAATCGGATTCGTCCCGCTGTGCCAGGAGAGCAGCGTGGTCCGCACTGT  
GTGCCTTCCCCCGGCGGACCTGCAGCTGCCGGACTIONGGACGGAGTGTGAGCTC

TCCGGCTACGGCAAGCATGAGGCCTTGTCTCCTTTCTATTTCGGAGCGGCTGA  
AGGAGGCTCATGTCAGACTGTACCCATCCAGCCGCTGCACATCACAACATTT  
ACTTAACAGAACAGTCACCGACAACATGCTGTGTGCTGGAGACACTCGGAGC  
GGCGGGCCCCAGGCAAACCTTGACGACGCCTGCCAGGGCGATTTCGGGAGGC  
CCCCTGGTGTGTCTGAACGATGGCCGCATGACTTTGGTGGGCATCATCAGCT  
GGGGCCTGGGCTGTGGACAGAAGGATGTCCCGGGTGTGTACACAAAGGTTA  
CCAACTACCTAGACTGGATTCGTGACAACATGCGACCGTGA (SEQ ID NO:7).

29. (Once amended) K2S protein according to claim 28 [or 29], characterized [characterised] in that it comprises a protein characterized by the following amino acid sequence or a fragment, a functional variant, an allelic variant, a subunit, a chemical derivative or a glycosylation variant thereof:

SEGNSDCYFGNGSAYRGTHSLTESGASCLPWNSMILIGKVYTAQNPSAQALGLG  
KHNYCRNPDGDAKPWCHVLKNRRLTWEYCDVPSCSTCGLRQYSQPQFRIKGGL  
FADIASHPWQAAIFAKHRRSPGERFLCGGILISSCWILSAAHCFQERFPPHHLTVIL  
GRTYRVVPGEEEQKFVEVEKYIVHKEFDDDTYDNDIALQLKSDSSRCAQESSVV  
RTVCLPPADLQLPDWTECELSGYGKHEALSPFYSERLKEAHVRLYPSSRCTSQHL  
LNRTVTDNMLCAGDTRSGGPQANLHDACQGDSSGGLVCLNDGRMTLVGIISWG  
LGCGQKDVPGVYTKVTNYLDWIRDNMRP\* (SEQ ID NO:11).

30. (Once amended) K2S according to [any one of claims] claim 27 [to 30], characterized [characterised] in that it consists of a protein characterized by the following amino acid sequence:

SEGNSDCYFGNGSAYRGTHSLTESGASCLPWNSMILIGKVYTAQNPSAQALGLG  
KHNYCRNPDGDAKPWCHVLKNRRLTWEYCDVPSCSTCGLRQYSQPQFRIKGGL  
FADIASHPWQAAIFAKHRRSPGERFLCGGILISSCWILSAAHCFQERFPPHHLTVIL  
GRTYRVVPGEEEQKFEVEKYIVHKEFDDDTYDNDIALQLKSDSSRCAQESSVV  
RTVCLPPADLQLPDWTECELSGYGKHEALSPFYSERLKEAHVRLYPSSRCTSQH  
LNRTVTDNMLCAGDTRSGGPQANLHDACQGDSGGPLVCLNDGRMTLVGIISWG  
LGCGQKDVPGVYTKVTNYLDWIRDNMRP\* (SEQ ID NO:11).

31. (Once amended) A vector containing a DNA sequence according to [any one of claims] claim 14 [to 24].

33. (Once amended) The vector pComb3HSS containing a DNA according to [any one of claims] claim 14 [to 24], wherein the expression of the gp III protein is suppressed or inhibited by deleting the DNA molecule encoding said gp III protein or by a stop codon between the gene coding for a polypeptide containing the kringle 2 domain and the serine protease domain of tissue plasminogen activator protein and the protein III gene.

34. (Once amended) A prokaryotic host cell comprising a DNA molecule according to [any one of claims] claim 14 [to 24].

35. (Once amended) A prokaryotic host cell comprising a vector according to [any one of claims] claim 31 [to 33].

36. (Once amended) An *E. coli* host cell comprising a DNA molecule according to [any one of claims] claim 14 [to 24].

37. (Once amended) An *E. coli* host cell comprising a vector according to [any one of claims] claim 31 [to 33].

Claims 38 and 39 were canceled.